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Seed origin drives differences in survival and growth traits of cork oak (*Quercus suber* L.) populations

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ABSTRACT

Cork oak (*Quercus suber* L.) has a high ecological and social value and supplies raw materials for the cork industry, a relevant contributor to the economies of Mediterranean countries. Understanding the adaptation potential of cork oak populations to cope with different environmental conditions is a key issue of forest management, particularly for selecting the most adapted genetic material for (re)forestation and assuring the long-term sustainability of the cork industry. Intraspecific variation in fitness surrogate traits (survival, height and stem diameter) was investigated in thirty-five cork oak populations sampled from the entire range of the natural distribution of the species. The study was conducted in two provenance field trials, established in Portugal under different edaphoclimatic conditions. Each trial was surveyed at four tree ages (two ages, 11 and 14 years, were sampled simultaneously in both trials). The trial located at a lower altitude, which had higher mean winter and annual temperatures, exhibited higher growth and survival rates. In both trials, significant genetic variation among cork oak populations was observed for the analyzed traits and evaluated ages. Moroccan populations displayed a higher probability of survival and higher growth rates, while local populations exhibited an intermediate performance. Low to moderate correlations were found between the analyzed traits and the environmental variables of seed origin, suggesting that factors other than climate are likely to be relevant for cork oak adaptation. Moderate to high values of population mean-basis broad-sense heritability ($H^2 \geq 0.44$) and high genetic correlations between traits (0.88–0.95) were found for growth traits. This information is crucial for the establishment of a breeding program for the species. With this study, we have improved the knowledge regarding how cork oak performs for fitness surrogate traits in different environments.

1. Introduction

Worldwide, forest ecosystems are facing increasing anthropogenic and environmental pressures from current and emerging threats that require a change in forest management paradigms. In the Mediterranean Basin, global warming is expected to have particularly adverse effects on tree survival, with the intensification of drought severity and an increase in pest and disease outbreaks (IPCC, 2014). Forest genetic resources can be used as a tool to cope with abiotic and biotic threats, as the use of well-adapted forest reproductive material will increase the competitive ability, survival and long-term success of tree species, contributing to the establishment of healthy and more productive forest stands.

Forest tree populations (also referred to as provenances) often show high levels of genetic variation (Wright, 1976), highlighting the importance of using the most suitable seed source behind the success of the (re)forestation programs. It is commonly assumed that local populations will have a higher chance of tolerating the conditions of the planting site. However, a rapidly changing environment introduces uncertainty in management and raises concerns that climate change is proceeding at a pace too fast for the adjustment by locally adapted trees (Higgins and Harte, 2006). Thus, the potential of tree species to endure rapid climate changes will depend on 'assisted migration' (Loss et al., 2011) and phenotypic plasticity and/or genetic adaptation (Valladares

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et al., 2007; Aitken et al., 2008; Alberto et al., 2013; Gratani, 2014). In this context, forest genetic trials, where provenances and/or progenies of different locations are grown in a common environment, represent a valuable resource to assess the pattern and levels of genetic variation among and/or within tree populations, how climate modulates phenotypes and whether this modulation is under genetic control. Thus, these trials represent a powerful tool for adapting forest management in response to climate change effects (Pâques, 2013).

Cork oak (*Quercus suber* L.) is a long-lived evergreen forest tree species with high socioeconomic and ecological importance for several western Mediterranean countries where the species occurs across a wide range of climatic and geographical conditions (Natividade, 1950; Aronson et al., 2009). The importance of this species is linked to its role as a source of income in rural areas and the provision of raw materials for the cork industry that depends on cork production and quality. Portugal hosts 34% of the world's cork oak area and accounts for 50% of the world's cork production (APCOR, 2018). However, since the 1980s, cork oak stands have been facing a significant vitality decline (Camilo-Alves et al., 2013) due to a combination of numerous factors, namely, a large proportion of old cork oak stands, poor natural regeneration and limited assisted regeneration, management (e.g excessive grazing intensity, root damage by heavy machinery) and low quality of forest reproductive material (seedlings with inadequate shoot/root ratio and low vigor) used on afforestation actions, more severe and frequent droughts that promote pest attacks and diseases, and the use of unsuitable genetic material for (re)forestation (e.g., Costa et al., 2010). This decline, and the consequent shortage of high-quality raw material, may be worsened by the environmental prospects foreseen for the Mediterranean Basin. Thus, the assessment of genetic variation of traits with adaptive and economic importance becomes essential to understanding the species' ability to cope with these pressures, assure the long-term sustainability of the cork industry, and deploy the most adapted genetic material for (re)forestation actions.

Previous studies highlighted significant variation in growth and morphological traits in cork oak population (Gandour et al., 2007) as well as in physiological traits mainly related to drought tolerance (Ramirez-Valiente et al., 2009a, 2009b, 2010, 2011, 2014a, 2014b, 2014c), and, in less extent, to cold resistance (Aranda et al., 2005). Additionally, in a cork oak field trial established in Portugal, significant differences among 35 populations were detected for vegetative phenology and leaf pest damage (Sampaio et al., 2016). Several of these studies have documented that the variation observed among cork oak populations in functional traits reflects a mechanism of local adaptation (Ramírez-Valiente et al., 2009a, 2010, 2014b), while population differences in plant architecture have been suggested to be associated with the past evolutionary history of the species (Ramírez-Valiente et al., 2014a). Additionally, it was reported that cork oak populations present high phenotypic plasticity for growth and leaf traits (Ramírez-Valiente et al., 2010, 2014b), i.e., populations can adapt to different environmental conditions by changing their morphological or physiological characteristics (Bradshaw, 1965), which constitutes an advantage for the species.

Despite the relevant findings on cork oak adaptive traits, most of the reported studies considered one single site and/or a small sample population, not allowing for a complete characterization of the natural distribution range of the species. Thus, local adaptation and phenotypic plasticity should be further investigated for fitness and functional traits considering several field trials and a wider climatic and geographic distribution range. In fact, in regions where this tree species occurs, water availability is the main factor limiting plant survival and growth (Larcher, 2000), especially in younger ages, before root development allows the tapping deeper water sources. Thus, understanding the survival and growth performance of cork oak trees in the first years is essential for the species' persistence. Additionally, stem growth is a key

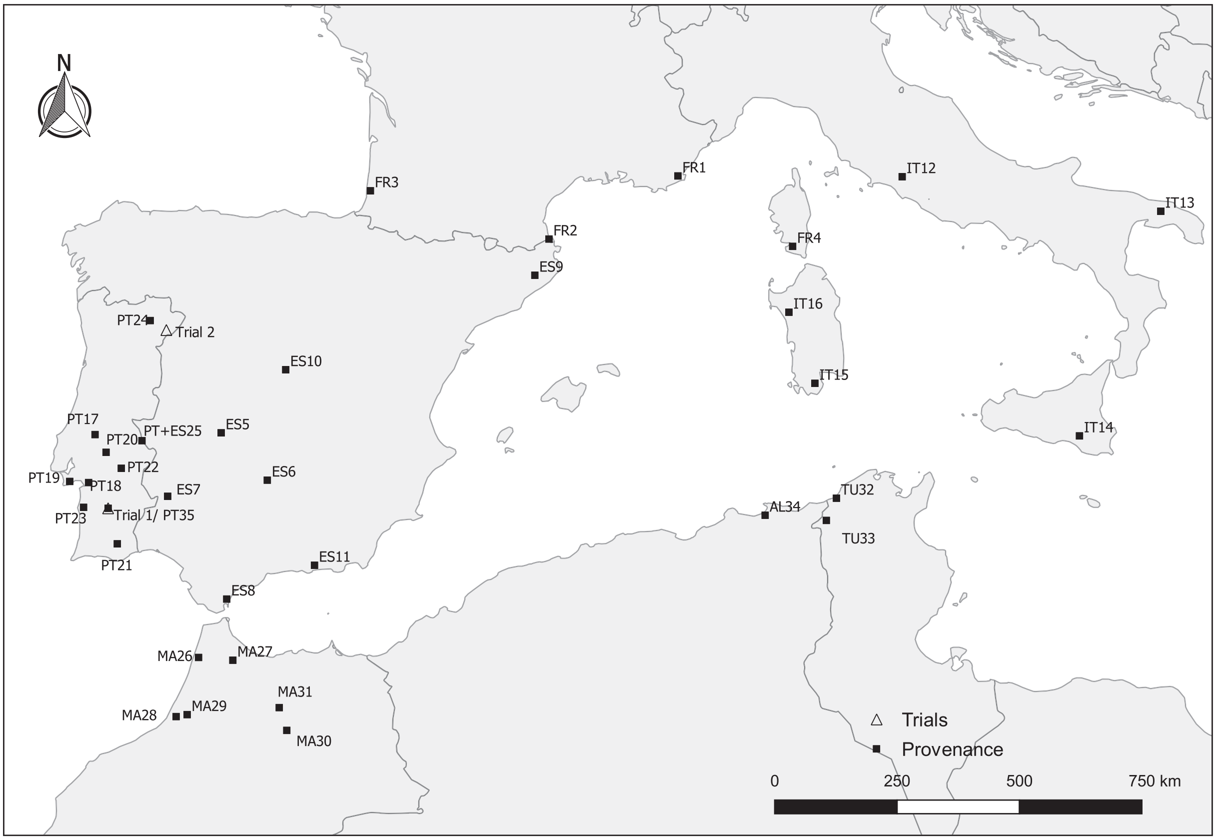

Fig. 1. Location of the sampled cork oak populations within the natural distribution range of the species and of the two environmentally contrasting cork oak provenance field trials in Portugal - Monte da Fava (Trial 1) and Quinta da Nogueira (Trial 2).

trait for this species, as cork (the most valuable non-wood forest product of cork oak stands) can only be stripped when the breast height perimeter reaches at least 70 cm (APCOR, 2018). In this sense, the main goal of this study was to evaluate the importance of adaptive mechanisms in fitness traits related to tree growth and survival to improve our understanding of the species potential for adaptation to different environments. Based on two different edaphoclimatic provenance field trials established in Portugal, where all the natural species distribution ranges are represented with 35 cork oak populations, the specific objectives of this study were (1) to assess the genetic variation at the population level for survival and growth traits (total height, root collar diameter and breast height diameter) under two different environmental conditions, (2) to identify populations with stable performance for those traits in those field trials and (3) to assess the population variation of the analyzed traits over time. As cork oak is a very important forest species both in Portugal and in the Western Mediterranean Basin and no superior material has been selected yet in the country, the expected knowledge gains arising from this study will allow to guide foresters in a possibility of delineate a breeding strategy for the species. Additionally, because of complete characterization of the species distribution range for relevant fitness traits, this study will be helpful to identify most-adapted seed sources to be used in (re)forestation practices and thus to contribute to more adapted and productive cork oak stands.

2. Materials and methods

2.1. Field trials and plant materials

The study was conducted using two Portuguese cork oak provenance field trials that are part of an international series of genetic trials, established under the framework of the Concerted Action, FAIR 1 CT 95-0202 (Varela, 2000). Both trials (Monte da Fava and Quinta da Nogueira) are characterized by a Mediterranean climate with hot, dry summers and cold and mild, wet winters, and precipitation is mainly concentrated in the October-May period. Plant material included in both field trials resulted from a seed collection conducted during the autumn of 1996 from 35 cork oak populations that cover the species' entire natural distribution range (Fig. 1). Detailed information on the location and climate data from both provenance trials and from the

Table 1

Location, and climate data (National Meteorological Agency) of the Monte da Fava and Quinta da Nogueira provenance trials. Information regarding the thirty-five cork oak populations represented in the two provenance trials. Code, location (closest locality, forest/estate and country) and geographic and climatic details for the populations' seed origin (adapted from Varela, 2000). Climate information for each population was collected at the closest weather station and provided by local Institutions, except for Algerian and Moroccan populations where data was obtained from a climate model (Hijmans et al., 2005). Included information is: Alt: altitude (m), P: precipitation (mm), sP: summer precipitation (mm), Tm: long-term mean annual air temperature (°C), Tsm: long-term mean summer air temperature (°C), Twm: long-term mean winter air temperature (°C), Tsmax: long-term mean air temperature for the hottest summer month (°C) and Twmin: long-term mean air temperature for the coldest winter month (°C). Country names were abbreviated as following: SP: Spain, IT: Italy, PT: Portugal, MA: Morocco, TU: Tunisia, AL: Algeria, Fr: France and PT-ES: border between Portugal and Spain.

Code	Location	Latitude	Longitude	Altitude	P	sP	T_m	T_{sm}	T_{wm}	T_{smax}	T_{wmin}
ES5	Cañamero, SP	39°23′ N	5°23′ W	700	1063	54	15.2	23.7	7.4	25.2	6.9
ES6	Fuencaliente, SP	38°28′ N	4°19′ W	800	719	43	14.3	22.8	5.6	24.7	4.4
ES7	Jerez de los Caballeros, El Carbajo, SP	38°13′ N	6°42′ W	450	666	34	16.0	23.9	8.7	25.2	8.1
ES8	Castellar de la Frontera, La Almoraima, SP	36°16′ N	5°22′ W	70	993	18	16.6	22.5	11.6	23.7	11.4
ES9	Santa Coloma de Farnés, SP	41°52′ N	2°35′ E	350	802	140	15.0	22.4	8.0	23.6	7.5
ES10	Madrid, El Pardo, SP	40°31′ N	3°45′ W	710	455	52	13.9	22.7	6.0	24.0	5.4
ES11	Haza de Lino, SP	36°50′ N	3°18 ' W	1300	742	26	13.0	20.2	6.9	21.9	6.3
IT12	Tuscania, Sughereta, IT	42°25′ N	11°57′ E	160	937	125	14.5	22.1	7.3	23.2	6.5
IT13	Brindisi, Lucci- S.Teresa, IT	40°34′ N	17°40′ E	45	588	49	16.6	23.7	9.9	24.6	9.1
IT14	Catania, Zotte, IT	37°07′ N	14°30′ E	250	448	9	17.7	25.1	11.4	26.2	11.1
IT15	Cagliari, Nuraghe Arcu de Mesu, IT	39°05′ N	8°51′ E	200	883	29	17.0	24.8	10.1	25.9	9.5
IT16	Sassari, Puttu addes de Subra, IT	40°27′ N	8°34′ E	300	910	43	14.2	22.0	7.2	22.9	6.6
PT17	Chamusca, Soc. Agrícola Igreja Velha, PT	39°23′ N	8°26′ W	75	829	46	15.6	21.9	9.5	22.9	9.1
PT18	Alcacer do Sal, Herdade da Palma, PT	38°29′ N	8°35′ W	30	577	26	16.3	22.2	10.7	23.0	10.3
PT19	Azeitão, Quinta da Serra, PT	38°30′ N	9°20′ W	120	681	19	14.3	19.0	9.7	19.7	9.4
PT20	Ponte de Sôr, Herdade de Vale Covo, PT	39°03′ N	8°10′ W	70	710	43	16.0	22.2	9.8	23.2	9.6
PT21	S. Brás de Alportel, PT	37°20′ N	7°54′ W	463	874	27	15.9	22.2	10.3	23.2	10.0
PT22	Azaruja, Herdade do Paço de Camões, PT	38°45′ N	7°48′ W	360	564	36	15.6	21.9	9.6	23.0	9.3
PT23	Santiago do Cacém, Monte Branco, PT	38°01′ N	8°42′ W	140	736	22	15.6	20.3	10.7	21.2	10.4
PT24	Romeu, Casa de Meneres, PT	41°31′ N	7°04′ W	456	666	57	14.2	20.1	5.2	31.6	2.0
PT35	Ermidas do Sado, Monte Fava, PT	38°00′ N	8°70′ W	79	557	19	15.8	22.0	10.4	31.3	4.3
MA26	Larache, Boussafi, MA	35°11′ N	6°03′ W	150	574	8	17.9	23.6	12.1	23.6	12.1
MA27	Chefchaouen, Ain Rami, MA	35°07′ N	5°16′ W	300	1280	26	17.0	24.2	10.4	24.2	10.4
MA28	Kenitra, Canton A, B, MA	34°05′ N	6°35′ W	160	536	9	18.1	23.7	12.5	23.7	12.5
MA29	Allal Baharoui, Ain Johra, MA	34°07′ N	6°20′ W	150	479	3	18.8	25.3	12.6	25.3	12.6
MA30	Oulmés, Oulmés, MA	33°46′ N	4°06′ W	1115	673	20	10.8	19.4	3.2	19.4	3.2
MA31	Taza, Bab Azhar, MA	34°12′ N	4°15′ W	1130	970	29	18.5	27.0	10.9	27.0	10.9
TU32	Ain Sobh, Tabarka, TU	36°57′ N	8°51′ W	12	948	27	17.9	24.3	11.6	25.6	11.1
TU33	Ain el Baya, Fernana, TU	36°35′ N	8°32′ E	270	1610	49	14.9	22.9	7.2	24.8	6.6
AL34	Guerbès, AL	36°90′ N	7°25′ E	45	760	26	17.4	24.0	11.0	24.0	11.0
FR1	Bornes les Mimosas, Les maures, FR	43°17′ N	6°30′ E	85	963	111	11.9	20.9	7.5	21.9	7.1
FR2	Collioure, Le Rimbault, FR	42°30′ N	3°03′ E	200	958	97	15.7	23.1	9.1	24.4	8.2
FR3	Soustons, FR	43°45′ N	1°20′ W	20	870	140	12.3	18.5	6.5	19.5	5.5
FR4	Sartene, FR	41°37′ N	8°58′ E	50	691	55	15.4	22.1	7.2	24.1	3.6
PTES25	Besteiros-Albuquerque, PT-ES	39°16′ N	7°18′ W	965	778	42	15.4	23.2	8.3	24.5	7.8
	Monte da Fava provenance trial – Long-term (1971–2000)	38°00′N	08°07′W	79	550	17	16.1	22.0	10.4	31.5	3.6
	Monte da Fava provenance trial – 2008	38°00′N	08°07′W	79	398	3	16.1	22.2	11.4	30.5	4.1
	Monte da Fava provenance trial – 2011	38°00′N	08°07′W	79	657	7	17.0	22.0	9.5	31.8	2.8
	Quinta da Nogueira provenance trial – Long-term (1971–2000)	41°20′N	06°40′W	754	562	67	12.2	20.1	5.2	21.9	0.0
	Quinta da Nogueira provenance trial – 2008	41°20′N	06°40′W	754	522	31	12.8	20.5	6.7	29.3	2.1
	Quinta da Nogueira provenance trial – 2011	41°20′N	06°40′W	754	523	29	14.2	21.0	6.4	30.0	2.7

tested plant material is given in Table 1. For each population, information includes code, country, location, altitude and climate data for seed sources. One of the French populations (coded as FR4) was not tested in the Quinta da Nogueira trial. Each seed lot was collected from at least 20 trees located 50–100 m apart. Mother tree selection was based on good acorn production and sanitary conditions. Acorns were sown in the same nursery for a one-year period, and seedlings were planted in field trials in 1998. The experimental design in each trial was a randomized complete block design with 30 blocks, each with two single-tree plot replicates per provenance. Trees were planted at 6 × 6 m spacing.

2.2. Phenotypic measurements: Survival and growth traits

Trait measurements included plant survival (S), total height (HT), root collar diameter (10 cm aboveground (DRC)), and breast height (130 cm) diameter (DBH). These traits were assessed in all plants of each field trial, i.e., 2 plants per population in each block at different tree ages. At Monte da Fava, survival and total height were assessed at ages 6, 9, 11, and 14 years, and root collar diameter was measured at ages 9, 11, and 14 years. At Quinta da Nogueira, survival and total height were assessed at ages 3, 8, 11, and 14 years, and root collar diameter was measured at ages 8, 11, and 14 years. DBH was assessed at both trials at age 14 years.

Traits were measured in autumn after the end of the vegetative season, which most frequently extends from spring to midsummer. Total height and diameter (root collar and breast height) were recorded in *m* and in *cm*, respectively, and survival was scored as *0* or *1* for dead and alive plants, respectively.

2.3. Data analysis

The theory of mixed models (McCulloch et al., 2008; Stroup et al., 2013) was used for statistical analyses.

2.3.1. Survival data

For survival data, a generalized linear mixed model was fitted for each site and age. The two binary observations on each block were considered. As a consequence, a model with a binomial distribution for response variable (number of surviving plants in 2 Bernoulli trials) and a logit link function was fitted. The considered model was as follows:

$$\boldsymbol{Y}^{*} = \boldsymbol{X\beta} + \boldsymbol{Zu},$$

where $\boldsymbol{Y}^{*}$ denotes the vector $n \times 1$ (in this case, $n = q_1 q_2$, whereq_1 is the number of blocks and q_2 is the number of populations), whose elements are given by $\ln\left(\frac{\pi_{ij}}{1-\pi_{ij}}\right)$ where π_{ij} is the probability of survival success for population j in the block i; $\boldsymbol{X}_{n\times p}$ is the design matrix for fixed effects; $\boldsymbol{\beta}_{p\times 1}$ is the vector of fixed effects (in this case, a scalar, the overall mean); $\boldsymbol{Z}_{n\times q}$ is the design matrix for random effects ($q = q_1 + q_2$); and $\boldsymbol{u}_{q\times 1}$ is the vector of random effects (includes the block and the population effects). Block and population effects were assumed to be independent and identically distributed normal random variables. That is, the covariance matrix $\boldsymbol{G}_{q\times q}$ was defined as $\boldsymbol{G} = \boldsymbol{G}_{block} \oplus \boldsymbol{G}_{pop}$, where $\boldsymbol{G}_{block} = \sigma^2_{block}\boldsymbol{I}_{q_1}$ (σ^2_{block} is the block variance, $\boldsymbol{I}_{q_1}$ is the identity matrix $q_1 \times q_1$) and $\boldsymbol{G}_{pop} = \sigma^2_{pop}\boldsymbol{I}_{q_2}$ (σ^2_{pop} is the population genetic variance, $\boldsymbol{I}_{q_2}$ is the identity matrix $q_2 \times q_2$).

The GLIMMIX procedure of SAS version 9.4 (SAS Institute Inc. 2015) was used to fit the generalized linear mixed model. Parameters were estimated via maximum likelihood by Laplace approximation.

2.3.2. Growth data

For growth trait data (total height and, root collar and breast height diameter), linear mixed models were fitted, which included single-site and across-site analyses. In matrix notation, the general formulation is described as:

$$\boldsymbol{Y} = \boldsymbol{X\beta} + \boldsymbol{Zu} + \boldsymbol{e}$$

where $\boldsymbol{Y}_{n\times 1}$ is the vector of observations (phenotypic values, which included the two single plot observations per population in each block), $\boldsymbol{X}_{n\times p}$ is the design matrix for fixed effects, $\boldsymbol{\beta}_{p\times 1}$ is the vector of fixed effects (includes the overall mean), $\boldsymbol{Z}_{n\times q}$ is the design matrix for random effects, $\boldsymbol{u}_{q\times 1}$ is the vector of random effects, and $\boldsymbol{e}_{n\times 1}$is the vector of random errors.

The vectors $\boldsymbol{u}$ and $\boldsymbol{e}$ are mutually independent, with multivariate normal distribution with vector of expected values $0_{q\times 1}$ and $0_{n\times 1}$ and covariance matrices $\boldsymbol{G}_{q\times q}$ and $\boldsymbol{R}_{n\times n}$, respectively. Consequently, the distribution of vector $\boldsymbol{Y}$ is multivariate normal, with an expectation of $\boldsymbol{X\beta}$ and covariance matrix $\boldsymbol{V} = \boldsymbol{ZGZ}^T + \boldsymbol{R}$, in which $\boldsymbol{Z}^T$ denotes the transpose of $\boldsymbol{Z}$.

2.3.2.1. Single-site analysis per age. In a first stage, linear mixed models considering block, population and block × population interaction as random effects factors were fitted for each growth trait and all ages per site. A nonsignificant block × population interaction was found for all analyzed traits at a given trial and age. This result was expected by the design of the experiment (only two single-tree plot replicates). Therefore, this term was not included in the final analysis. Block, population and random errors were assumed to be independent and identically distributed normal random variables. That is, the covariance matrix $\boldsymbol{G}_{q\times q}$ was defined as $\boldsymbol{G} = \boldsymbol{G}_{block} \oplus \boldsymbol{G}_{pop}$, where $\boldsymbol{G}_{block} = \sigma^2_{block}\boldsymbol{I}_{q_1}$ (σ^2_{block} is the block variance, $\boldsymbol{I}_{q_1}$ is the identity matrix $q_1 \times q_1$, and q_1 is the number of blocks) and $\boldsymbol{G}_{pop} = \sigma^2_{pop}\boldsymbol{I}_{q_2}$ (σ^2_{pop} is the population genetic variance, $\boldsymbol{I}_{q_2}$ is the identity matrix $q_2 \times q_2$, and q_2 is the number of populations). The covariance matrix of random errors was defined as $\boldsymbol{R} = \sigma^2_e\boldsymbol{I}_n$ (σ^2_e is the error variance, $\boldsymbol{I}_n$ is the identity matrix $n \times n$, and n is the number of observations).

Additionally, to evaluate the population genetic correlation between growth traits (i.e., trait-trait correlations), a multitrait analysis was conducted at each site for ages 11 and 14 years. In this case, the vector of observations was defined as $\boldsymbol{Y} = (\boldsymbol{Y}_1^{\boldsymbol{T}}, \boldsymbol{Y}_2^{\boldsymbol{T}}, \cdots, \boldsymbol{Y}_{\boldsymbol{t}}^{\boldsymbol{T}})^{\boldsymbol{T}}$, comprised of t subvectors, each corresponding to each growth trait. The vector of fixed effects included the overall means for each trait, and the vector of random effects included the effects of the blocks within the trait and the population effects for each trait. The covariance matrix for block effects was defined as $\boldsymbol{G}_{block} = \sum_{block} \otimes \ \boldsymbol{I}_{q_1}$, where $\sum_{block}$ was a diagonal matrix $t \times t$ with diagonal elements given by $\sigma^2_{block_j}$ (block variance for trait j, for $j = 1, \ \cdots, t$), and $\otimes$ is the Kronecker product. The covariance matrix for population effects was defined as $\boldsymbol{G}_{pop} = \sum_{pop} \otimes \ \boldsymbol{I}_{q_2}$, where $\sum_{pop}$ is an unstructured covariance matrix $t \times t$, with diagonal elements given by $\sigma^2_{pop_j}$ (population genetic variance for trait j, for $j = 1, \ \cdots, t$), and nondiagonal elements given by $\sigma_{pop_j pop_{j}^{'}}$ (population covariance between traits j and j', $\forall\, j \neq j^{'}$). The population genetic correlation between traits j and j' ($\rho_{pop_j pop_{j'}}$) was calculated as: $\rho_{pop_j pop_{j'}} = \frac{\sigma_{pop_j pop_{j'}}}{\sigma_{pop_j}\sigma_{pop_{j'}}}$. An analogous covariance structure for random errors was adopted.

2.3.2.2. Single-site analysis including all studied ages. A single-site analysis including all studied ages was conducted to understand the performance of each population across ages and to evaluate the population × age interaction. Considering the general linear mixed model described above, the vector of the fixed effects included the overall mean as well as age effects, and the vector of random effects included blocks within age, population and population by age interaction effects. All random effects were assumed to be independent and identically distributed normal random variables. Random errors were assumed to be normal random variables, and an error covariance matrix incorporating correlations for all the observations arising from the same tree was considered. Specifically, a power correlation function was used.

2.3.2.3. Across-site analysis. To assess the population × site interaction at ages 11 and 14 years, two approaches were conducted. A classical analysis to estimate the population × site interaction variance components and to test its significance. Considering the general linear mixed model described above, the vector of the fixed effects included the overall mean as well as site effects, and the vector of random effects included blocks within site, population and population × site interaction effects. All random effects and random errors were assumed to be independent and identically distributed normal random variables. An across-site analysis considering heterogeneous variances and non-diagonal covariance matrices was also conducted to evaluate the population genetic correlation between sites (another measure to study population × site interaction) at age 11 and 14 years. In this way, the causes of population × site interaction are separated: the interaction due to scale (heterogeneity of population variation between sites) and the interaction due to crossover which affects the ranking of populations (population genetic correlation between sites). In the linear mixed model, the vector of the fixed effects included the overall mean as well as site effects. The vector of random effects included blocks within site and populations within site effects. Heterogeneous variances between sites were assumed for blocks within site effects, for populations within site effects and for random errors within site effects. Specifically, the covariance matrix for blocks within site effects was defined as $\boldsymbol{G}_{block\,(site)} = \sum_{block\,(site)} \otimes \boldsymbol{I}_{q_1}$, where $\sum_{block\,(site)}$ is a diagonal matrix 2×2 with diagonal elements given by $\sigma^2_{block_j}$ (block variance for site j, for $j = 1, 2$), and $\otimes$ is the Kronecker product. The covariance matrix for population within site effects was defined as $\boldsymbol{G}_{pop\,(site)} = \sum_{pop\,(site)} \otimes \boldsymbol{I}_{q_2}$, where $\sum_{pop\,(site)}$ is an unstructured covariance matrix 2×2, with diagonal elements given by $\sigma^2_{pop_j}$ (population genetic variance for site j, for $j = 1, 2$), and nondiagonal elements given by $\sigma_{pop_1pop_2}$ (population covariance between site 1 and 2). The population genetic correlation between sites 1 and 2 ($\rho_{pop_1pop_2}$) was calculated as: $\rho_{pop_1pop_2} = \frac{\sigma_{pop_1pop_2}}{\sigma_{pop_1}\sigma_{pop_2}}$. The higher the positive correlation between sites, the lower the population × site interaction. A similar covariance structure for random errors was adopted.

The ASREML-R package (Butler et al., 2007) was used to fit the linear mixed models described for single-site and across-site analyses. The variance-covariance parameters were estimated by the residual maximum likelihood (REML) method. Empirical best linear unbiased estimators (EBLUEs) of the fixed effects and empirical best linear unbiased predictors (EBLUPs) of the random effects were obtained from mixed model equations (Henderson, 1975).

For survival and growth models, covariance components were tested using likelihood ratio tests. The asymptotic distribution of the likelihood ratio test statistic was assumed to be a chi-squared distribution with the number of degrees of freedom equal to the increase in the number of parameters between the two compared models. A Wald-type F test was used to test growth differences between site means. All analyses were performed at a significance level of 5%.

All model assumptions were validated by graphical diagnostic tools for the residuals and for the empirical best linear unbiased predictors of random effects.

2.3.3. Genetic parameters and population performance

To study the survival population performance, the mean predicted survival percentage of each population was ranked. The mean predicted survival percentage for population j ($\bar{\hat{\pi}}_{.j}$) was calculated as:

$$\bar{\hat{\pi}}_{.j}=\left(\frac{\sum_{i=1}^{q_1}\hat{\pi}_{ij}}{q_1}\right)\times 100$$

where $\hat{\pi}_{ij}$, the predicted survival for population j in the block i, was obtained from the inverse of the link function as:

$$\hat{\pi}_{ij} = \frac{1}{1 + e^{[-EBLUE\,(overallmean)-\left(EBLUP\,(block_i)+EBLUP\,(pop_j)\right)]}}$$

For growth traits, population performance was studied ranking the EBLUPs of the population effects obtained from the across-site analysis with heterogeneous variances.

A generalized measure of population mean-basis broad-sense heritability (H^2) (adapted from Oakey et al., 2006) was calculated for growth traits:

$$H^2 = \frac{\sum_{j=1}^{q_2}\left(1 - \frac{\widehat{PEV}_j}{\hat{\sigma}^2_{pop}}\right)}{q_2 - 1},$$

where $\widehat{PEV}_j$ is the prediction error variance estimate of the effect for population j ($j = 1, \cdots, q_2$), and $\hat{\sigma}^2_{pop}$ is the variance component estimate for the population effects.

2.3.4. Correlation between survival and growth traits and the environmental variables of the population's site of origin

Nonlinear trends were not observed between traits (mean predicted percentage of survival and predicted values of total height and aboveground diameter) and the geographical and climatic parameters of the population's site of origin, at age 14 years at both field trials. Thus, Pearson's correlation coefficients were used to obtain the correlation between these traits. For this analysis, the CORR procedure of SAS version 9.4 (SAS Institute Inc. 2015) was used.

3. Results

3.1. Survival data

The results for survival data are shown in Table 2. Significant population variance estimates ($p < 0.05$) were observed at both sites and all evaluated ages, except for ages 3 and 8 years, at Quinta da Nogueira. Regarding the common evaluated ages at both sites (11 and 14 years), higher predicted survival rate means were observed at Monte da Fava. The mean predicted survival rate was consistently greater than the years at Monte da Fava, while at Quinta da Nogueira, a decrease was observed. At Monte da Fava, a slightly higher predicted survival rate was observed for ages 9, 11 and 14 compared with that of age 6 years because cork oak has the capacity to resprout. High survival

Table 2

Predicted mean percentage of population survival ($\hat{\pi}$) and estimates of variance components obtained under the generalized linear mixed model (in logit scale) for survival, at different ages and for the test sites Monte da Fava and Quinta da Nogueira; the respective *p-values* for the variance components are given in parentheses.

Site	Age	$\hat{\pi}$	Variance	
			Population	Block
Monte da Fava	6	84.4	0.155 ($p < 0.001$)	1.273 ($p < 0.001$)
	9	84.9	0.096 ($p = 0.020$)	1.223 ($p < 0.001$)
	11	84.9	0.121 ($p = 0.005$)	1.366 ($p < 0.001$)
	14	84.8	0.103 ($p = 0.015$)	1.314 ($p < 0.001$)
Quinta da Nogueira	3	96.4	0	0.112 ($p = 0.332$)
	8	75.2	0.015 ($p = 0.603$)	1.953 ($p < 0.001$)
	11	73.9	0.068 ($p = 0.041$)	1.941 ($p < 0.001$)
	14	69.6	0.068 ($p = 0.021$)	1.489 ($p < 0.001$)

At each site and for the respective evaluated ages, Pearson χ^2/df statistics values, ranged between 0.79 and 0.91, which indicates that there is no evidence of overdispersion.

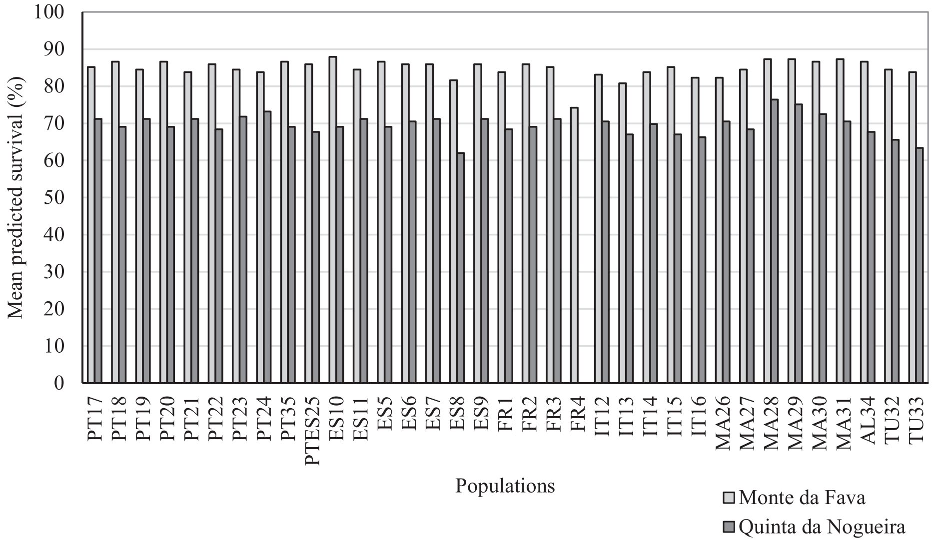

Fig. 2. Survival performance for the sampled populations, at the test sites Monte da Fava and Quinta da Nogueira, at age 14 years old.

performance was observed for the sampled populations, at both test sites at age 14 years old (Fig. 2). Despite the range of predicted survival rate means was small, it can be highlighted that the Spanish population from La Almoraima (ES8) was at the bottom ranked position at both sites and, the French population from Sertene (FR4) and the Italian population from Brindisi (IT13) presented the lowest survival performance at Monte da Fava. The more occidental Moroccan populations (MA28 and MA29) were among those populations with higher mean predicted survival rates at both sites. However, at Monte da Fava, the population from Madrid (ES10) was top-ranked, whereas at Quinta da Nogueira, this population was intermediately ranked. The Portuguese populations from Santiago do Cacém and Romeu (PT23 and PT24, respectively) were among at the top of survival performance at Quinta da Nogueira (see also Table S1 in the supplementary material).

Supplementary data associated with this article can be found, in the online version, at https://doi.org/10.1016/j.foreco.2019.05.001.

3.2. *Growth traits*

The results for growth traits obtained from the single-site analysis per age are given in Table 3. At both sites and for the respective evaluated ages, highly significant population effects ($p < 0.001$) were observed. A more effective genetic control (higher values of broad-sense heritability) of growth traits was observed at Monte da Fava. The heritability values increased with age and varied between 0.44 (for DRC at age 8 years at Quinta da Nogueira) and 0.88 (for HT at age 14 years at Monte da Fava).

Population genetic correlations estimates between growth (HT and DRC) traits obtained from the fitting of the multitrait model to each site at ages 11 and 14 years were, as expected, high and positive. These significant ($p < 0.001$) estimates, were 0.88 and 0.95 at Quinta da Nogueira for age 11 and 14 years, respectively. At Monte da Fava this correlation was 0.94 for both evaluated ages.

The results of the single-site analysis including all ages are shown in Table 4. Across ages, correlations between observations from the same

Table 3
Estimates of variance components obtained under the single-site analyses and values of population mean-basis broad-sense heritability (H^2), for growth traits (total height – HT, root collar diameter – DRC, and breast height diameter – DBH), at different ages for the test sites Monte da Fava and Quinta da Nogueira; the respective *p-values* for the variance components are given in parentheses.

Age	Trait	Monte da Fava				Age	Trait	Quinta da Nogueira			
		Variance			H^2			Variance			H^2
		Population	Block	Error				Population	Block	Error	
6	HT	0.016 ($p < 0.001$)	0.075 ($p < 0.001$)	0.221	0.775	3	HT	0.001 ($p < 0.001$)	0.003 ($p < 0.001$)	0.041	0.624
9	HT	0.029 ($p < 0.001$)	0.143 ($p < 0.001$)	0.378	0.781	8	HT	0.010 ($p < 0.001$)	0.028 ($p < 0.001$)	0.231	0.665
	DRC	0.440 ($p < 0.001$)	2.534 ($p < 0.001$)	5.765	0.779		DRC	0.056 ($p < 0.01$)	0.527 ($p < 0.001$)	3.212	0.444
11	HT	0.109 ($p < 0.001$)	0.417 ($p < 0.001$)	0.978	0.839	11	HT	0.039 ($p < 0.001$)	0.108 ($p < 0.001$)	0.762	0.685
	DRC	1.149 ($p < 0.001$)	5.757 ($p < 0.001$)	12.464	0.812		DRC	0.248 ($p < 0.001$)	1.044 ($p < 0.001$)	7.699	0.581
14	HT	0.245 ($p < 0.001$)	0.892 ($p < 0.001$)	1.530	0.882	14	HT	0.060 ($p < 0.001$)	0.143 ($p < 0.001$)	1.132	0.680
	DRC	2.361 ($p < 0.001$)	7.750 ($p < 0.001$)	17.770	0.861		DRC	0.535 ($p < 0.001$)	1.696 ($p < 0.001$)	11.303	0.657
	DBH	1.801 ($p < 0.001$)	3.926 ($p < 0.001$)	13.580	0.854		DBH	0.609 ($p < 0.001$)	1.383 ($p < 0.001$)	9.960	0.692

Table 4

Estimates of covariance components obtained from the single-site analysis including all ages, autocorrelation parameter estimates (ϕ) and values of population mean-basis broad-sense heritability (H^2), for growth traits (total height – HT, and root collar diameter – DRC) for the test sites Monte da Fava and Quinta da Nogueira; the respective *p-values* for the variance components are given in parentheses.

Site	Trait	Population	Population × Age	Block(Age)	Error	ϕ	H^2
Monte da Fava	HT	0.071 ($p < 0.001$)	0.021 ($p < 0.001$)	0.356 ($p < 0.001$)	0.786	0.898	0.917
	DRC	1.150 ($p < 0.001$)	0.160 ($p < 0.001$)	4.833 ($p < 0.001$)	12.353	0.958	0.827
Quinta da Nogueira	HT	0.015 ($p < 0.001$)	0.007 ($p < 0.001$)	0.052 ($p < 0.001$)	0.447	0.918	0.702
	DRC	0.216 ($p < 0.001$)	0.050 ($p < 0.001$)	0.761 ($p < 0.001$)	7.200	0.950	0.595

The correlation between two ages is the given by ϕ^u, where u is the lag distance between ages.

tree were similar in both sites and were higher for root collar diameter. Significant population × age interaction was found for both traits. Moderate to high values of population mean-basis broad-sense heritability were obtained.

The results of across-site analyses considering the classical population × site interaction approach for growth traits at ages 11 and 14 years old are given in Table 5. Significant population × site interaction variance ($p < 0.05$) was found for all cases. The results considering the multienvironment analyses with heterogeneous variances are shown in Tables 6 and 7. Significant differences between site means ($p < 0.001$) were observed for all growth traits. For ages 11 and 14 years old, the mean height was higher at Monte da Fava than that at Quinta da Nogueira. The same pattern was observed for DRC and DBH (Table 6). The broad-sense heritability values obtained with this analysis varied between 0.64 (for DRC at age 11 years at Quinta da Nogueira) and 0.91 (for HT at age 14 years at Monte da Fava).

The across-site correlation for population effects (the part of population × site interaction due to the change on the populations ranking) varied between 0.60 (for DRC at age 11 years) and 0.87 (for HT at age 11 years) (Table 6). This range of correlations also confirmed some degree of population × site interaction, but at a moderate to low level, as seen by the ranking of the EBLUPs of population effects at age 14 years (Table 7). In general, the population rankings were consistent across sites and traits. Moroccan populations displayed consistently high performance for height and, root collar and breast height diameters at both sites (Table 7). In particular, the more occidental Moroccan populations (MA28 and MA29) and the populations from Oulmés (MA30) and Ain Rami (MA27) were among those top-ranked for all studied growth traits. On the opposite extreme, the populations from Brindisi (IT13) and from Haza de Lino (ES11) ranked consistently among the bottom five populations. French populations (FR2, FR3 and FR4) consistently exhibited poor growth performance. The Spanish population from Fuencaliente (ES6) also displayed a poor performance, especially at Quinta da Nogueira. Portuguese populations occupied an intermediate place at both sites.

Table 5

Estimates of variance components obtained from the across-site analysis considering the classical approach and, values of population mean-basis broad-sense heritability (H^2), for growth traits (total height – HT, root collar diameter – DRC, and breast height diameter – DBH) at age 11 and 14 years; the respective *p-values* for the variance components are given in parentheses.

Age	Trait	Population	Population × Site	Block(Site)	Error	H^2
11	HT	0.057 ($p = 0.002$)	0.018 ($p = 0.012$)	0.262 ($p < 0.001$)	0.879	0.843
	DRC	0.342 ($p = 0.046$)	0.371 ($p = 0.001$)	3.389 ($p < 0.001$)	10.246	0.711
14	HT	0.098 ($p = 0.007$)	0.058 ($p < 0.001$)	0.515 ($p < 0.001$)	1.352	0.846
	DRC	0.921 ($p = 0.014$)	0.581 ($p < 0.001$)	4.723 ($p < 0.001$)	14.859	0.827
	DBH	0.915 ($p = 0.003$)	0.336 ($p = 0.004$)	2.684 ($p < 0.001$)	11.970	0.849

3.3. Correlation between the evaluated traits and the environmental and geographic parameters of the origin site

Pearson's correlation coefficients for survival and growth traits and geographic and environmental parameters of the site of the population's origin are given in Table 8. At Monte da Fava, negative and moderate correlations were found between growth traits, latitude and climatic variables (total precipitation of the summer months). Additionally, a low-moderate, positive correlation between growth traits and mean temperature, winter mean temperature and winter mean temperature of the coldest month was observed at this site. At Quinta da Nogueira, the effect of latitude remained significant at a moderate level for growth traits, but longitude was only significantly low when correlated with HT. Summer precipitation of the population seed source was low and negatively correlated with DRC at Quinta da Nogueira. Survival data were only statistically correlated with longitude at a moderate level at Monte da Fava and with precipitation at Quinta da Nogueira.

4. Discussion

In this study, significant population variation for fitness surrogate traits (survival, height and, root collar and breast height diameter growth) was detected across cork oak's natural distribution, which agrees with previous studies in which these traits were also analyzed (Gandour et al., 2007; Ramírez-Valiente et al., 2009b, 2014b). The existence of interpopulation variation shows that cork oak populations do not perform similarly with regard to fitness traits. Thus, it will be essential to use the most adapted material for (re)forestation to maximize the species productivity and contribute to its long-term maintenance under ongoing environmental changes. Additionally, models anticipating climate change-driven tree redistribution for the Mediterranean region (e.g., Benito Garzón et al., 2008; Vessella et al., 2017; Duque-Lazo et al., 2018) should start considering within-species variation for a more efficient management and conservation of the forest resources. An important advance in these models was the work of Schirone et al. (2016) where ecological niche modeling was performed according to different climatic scenarios to forecast the expansion/retreat of 4 cork oak haplotypes. Coupled with an understanding of cork oak evolutionary history (Schirone et al., 2015), this study opens the possibility of an assisted migration strategy for the species under ongoing environmental changes.

In a previous study conducted in the Monte da Fava provenance trial, high values of heritability ($H^2 = 0.79–0.94$) were found for vegetative phenology (Sampaio et al., 2016). Similarly, in the current study high values of broad-sense heritability at the level of population means were obtained for growth traits at both sites, suggesting an

Table 6
Overall mean estimates, variance components estimates, values of population mean-basis broad-sense heritability (H^2), mean differences between sites and, population correlation between sites, obtained from the across-site analyses (with heterogeneous variances and non-diagonal covariance matrices) for growth traits (total height – HT, root collar diameter – DRC, and breast height diameter – DBH, at age 11 and 14 years; Overall mean estimates are in *m* for HT and in *cm* for DRC and DBH; the respective *p-values* for the variance components and population correlation between sites are given in parentheses.

		Monte da Fava					Quinta da Nogueira					Between sites	
		Mean	Variance			H^2	Mean	Variance			H^2	Mean differences	Population correlation
Trait	Age		Population	Block	Error			Population	Block	Error			
HT	11	3.164 ± 0.136	0.109 ($p < 0.001$)	0.418 ($p < 0.001$)	0.978	0.875	2.456 ± 0.073	0.038 ($p < 0.001$)	0.108 ($p < 0.001$)	0.765	0.821	$p < 0.001$	0.870 ($p < 0.001$)
	14	4.549 ± 0.200	0.245 ($p < 0.001$)	0.893 ($p < 0.001$)	1.530	0.908	2.888 ± 0.087	0.058 ($p < 0.001$)	0.145 ($p < 0.001$)	1.136	0.795	$p < 0.001$	0.796 ($p < 0.001$)
DRC	11	10.784 ± 0.496	1.148 ($p < 0.001$)	5.760 ($p < 0.001$)	12.465	0.831	7.972 ± 0.222	0.242 ($p < 0.001$)	1.072 ($p < 0.001$)	7.701	0.644	$p < 0.001$	0.604 ($p < 0.001$)
	14	14.178 ± 0.597	2.361 ($p < 0.001$)	7.764 ($p < 0.001$)	17.769	0.888	10.356 ± 0.290	0.532 ($p < 0.001$)	1.742 ($p < 0.001$)	11.336	0.763	$p < 0.001$	0.757 ($p < 0.001$)
DBH	14	9.951 ± 0.449	1.809 ($p < 0.001$)	3.937 ($p < 0.001$)	13.580	0.890	6.571 ± 0.275	0.598 ($p < 0.001$)	1.446 ($p < 0.001$)	9.954	0.815	$p < 0.001$	0.837 ($p < 0.001$)

effective genetic selection and a possibility to start a breeding program for cork oak. The increase of broad-sense heritability with age, is usually considered a consequence of the competition effect on growth (Foster, 1989). However, our results reflect a higher genetic difference among populations over time rather than a competitive situation, as the studied trees were in the juvenile stage (14 years), and the space between them was still large. In general, the high values of broad-sense heritability at the level of population means obtained in this study, can

Table 7
Empirical best linear unbiased predictors (EBLUPs) of population effects for growth traits (total height - HT, root collar diameter - DRC, and breast height diameter - DBH) at age 14 years, ordered from the largest to smallest, at the test sites Monte da Fava and Quinta da Nogueira, obtained from across-site analysis (with heterogeneous variances).

HT				DRC				DBH			
Monte da Fava		Quinta da Nogueira		Monte da Fava		Quinta da Nogueira		Monte da Fava		Quinta da Nogueira	
Population	EBLUP	Population	EBLUP	Population	EBLUP	Population	EBLUP	Population	EBLUP	Population	EBLUP
MA28	1.134	MA30	0.491	MA28	3.139	MA30	1.311	MA28	3.079	MA28	1.407
MA29	1.104	MA28	0.463	MA29	2.964	MA28	1.178	MA26	2.724	MA30	1.382
MA27	0.809	MA29	0.357	MA26	2.892	MA29	0.887	MA29	2.032	MA27	0.995
MA26	0.798	MA27	0.299	MA27	1.930	MA27	0.886	MA27	1.794	MA26	0.930
MA30	0.702	ES8	0.218	MA30	1.613	MA31	0.809	MA30	1.770	ES8	0.693
AL34	0.275	MA26	0.191	IT12	0.748	ES8	0.687	MA31	0.872	MA29	0.686
MA31	0.220	MA31	0.166	PT35	0.739	MA26	0.586	PT17	0.867	MA31	0.646
PT23	0.190	AL34	0.140	AL34	0.581	ES7	0.396	PT20	0.625	PT20	0.441
ES8	0.175	ES7	0.125	MA31	0.568	PT20	0.375	ES8	0.608	ES7	0.421
PT20	0.124	PT20	0.118	PT23	0.563	AL34	0.360	PT18	0.489	PT17	0.413
PT17	0.102	PT22	0.082	ES8	0.552	PT35	0.331	PT23	0.437	PT22	0.317
PT21	0.062	PT24	0.040	PT18	0.528	PT18	0.172	ES7	0.393	PT18	0.295
PT22	0.035	ES5	0.036	PT20	0.490	IT16	0.168	PT21	0.376	ES5	0.266
PT35	0.033	PT18	0.032	TU33	0.419	PT22	0.100	AL34	0.247	AL34	0.221
PT24	0.023	PT23	0.026	IT16	0.378	TU33	0.090	PT35	0.164	PT23	0.175
PT18	0.021	PT35	0.025	IT14	0.293	PT23	0.062	PT22	0.059	PT21	0.146
PTES25	−0.027	FR1	−0.002	PT21	0.219	IT14	−0.008	TU33	−0.003	PT35	0.090
IT12	−0.052	TU33	−0.024	PT17	0.209	FR1	−0.014	IT12	−0.127	IT16	−0.013
TU33	−0.053	TU32	−0.042	PT22	0.113	IT12	−0.018	PTES25	−0.132	TU33	−0.097
PT19	−0.070	ES9	−0.055	PT19	0.071	PT21	−0.046	ES5	−0.136	PTES25	−0.212
ES7	−0.088	PT17	−0.055	ES7	−0.154	ES5	−0.054	PT19	−0.197	FR1	−0.227
ES5	−0.107	PT21	−0.064	PTES25	−0.249	PT19	−0.225	IT14	−0.243	PT19	−0.230
TU32	−0.122	PTES25	−0.090	TU32	−0.311	PTES25	−0.256	IT16	−0.248	PT24	−0.261
FR1	−0.130	IT16	−0.091	ES5	−0.515	TU32	−0.258	TU32	−0.518	ES9	−0.273
IT16	−0.174	PT19	−0.116	FR1	−0.759	PT17	−0.281	PT24	−0.561	TU32	−0.277
IT14	−0.225	IT12	−0.128	PT24	−0.780	ES10	−0.309	FR1	−0.743	IT14	−0.360
ES6	−0.318	IT14	−0.140	IT15	−0.845	ES9	−0.360	IT15	−0.848	IT12	−0.430
FR3	−0.338	ES10	−0.147	ES6	−1.480	PT24	−0.385	ES9	−1.102	ES10	−0.434
FR4	−0.414	FR2	−0.170	ES10	−1.606	FR2	−0.484	ES6	−1.347	FR2	−0.584
IT15	−0.463	FR3	−0.204	ES9	−1.653	IT15	−0.556	FR3	−1.482	IT15	−0.604
ES10	−0.486	ES6	−0.231	FR3	−1.717	IT13	−1.006	ES10	−1.491	ES6	−0.945
ES9	−0.492	IT15	−0.265	IT13	−1.866	ES6	−1.018	FR2	−1.638	FR3	−1.002
ES11	−0.660	ES11	−0.388	ES11	−2.122	ES11	−1.061	IT13	−1.811	ES11	−1.253
FR2	−0.740	IT13	−0.435	FR2	−2.185	FR3	−1.063	ES11	−1.942	IT13	−1.377
IT13	−0.849			FR4	−2.768			FR4	−1.965		

Table 8

Pearson's correlation coefficients and respective *p-values* (in parentheses) between mean predicted percentage of survival (S), predicted total height (HT), and predicted root collar diameter (DRC) at age 14 years at both test sites and geographic and environmental variables of the population's origin site: latitude (Lat), longitude (Long), altitude (Alt, m), precipitation (P, mm), summer precipitation (sP, mm); long-term mean annual air temperature (Tm, °C), long-term mean summer air temperature (Tsm, °C), long-term mean winter air temperature (Twm, °C), long-term mean air temperature for the hottest summer month (Tsmax. °C) and long-term mean air temperature for the coldest winter month (Twmin, °C). N = 35. Bold values are statistically significant ($p < 0.05$).

Trial	Age	Trait	Lat	Long	Alt	P	sP	Tm	Tsm	Twm	Tsmax	Twmin
Monte da Fava	14	S	−0.274	**−0.464**	0.303	−0.122	−0.087	0.050	0.141	0.072	0.051	0.179
			(0.111)	(**0.005**)	(0.077)	(0.487)	(0.621)	(0.776)	(0.419)	(0.682)	(0.772)	(0.303)
		HT	**−0.703**	**−0.444**	−0.078	−0.081	**−0.500**	**0.367**	0.174	**0.411**	−0.035	**0.441**
			(< **0.001**)	(**0.008**)	(0.656)	(0.645)	(**0.002**)	(**0.030**)	(0.318)	(**0.014**)	(0.843)	(**0.008**)
		DRC	**−0.702**	−0.327	−0.166	−0.081	**−0.534**	**0.444**	0.245	**0.520**	0.000	**0.547**
			(< **0.001**)	(0.055)	(0.340)	(0.644)	(**0.001**)	(**0.008**)	(0.157)	(**0.001**)	(0.998)	(**0.001**)
Quinta da Nogueira	14	S	−0.176	−0.323	0.158	**−0.491**	−0.033	0.005	−0.105	−0.027	−0.08	−0.008
			(0.318)	(0.063)	(0.373)	(**0.003**)	(0.853)	(0.977)	(0.555)	(0.881)	(0.643)	(0.965)
		HT	**−0.509**	**−0.389**	0.042	−0.010	−0.261	0.139	0.130	0.119	0.028	0.161
			(**0.002**)	(**0.023**)	(0.814)	(0.957)	(0.137)	(0.432)	(0.463)	(0.504)	(0.877)	(0.363)
		DRC	**−0.589**	−0.274	0.089	0.021	**−0.373**	0.212	0.259	0.210	0.033	0.253
			(< **0.001**)	(0.117)	(0.618)	(0.905)	(**0.030**)	(0.229)	(0.139)	(0.233)	(0.852)	(0.148)

be justified by the experimental design of the trials, concretely by the number of replicates (two single-tree plots per block). Additionally, the higher the complexity of the model, the higher the heritability values obtained. This increase is due to the decrease of the prediction error variance estimate associated to the EBLUPs of the effects of the population in the more complex models, namely in single-site analysis including all evaluated ages and across-site analysis with heterogeneous variances and non-diagonal covariance matrices.

The high genetic correlations observed between growth traits at different ages indicate that, from a breeding perspective, it will be preferable to select for cork oak diameter rather than tree height because it is more relevant as it is linked with increased productivity (extraction of wider cork planks). Furthermore, it is easier to measure and allows for fewer experimental errors. Our study also revealed higher diameter correlations across ages, which reflects the lower diameter increments between years compared with the height increments, for the same period. The significant population × age interaction revealed that not all populations showed a similar growth behavior for growth traits over time.

The significant differences observed between site means seems to be related to edaphoclimatic characteristics. Several and coupled reasons can have contributed for the lower performance observed at Quinta da Nogueira: i) the higher altitude, where the added effects of a shorter growing season and lower temperatures can affect the phenological window and consequently, plant growth; ii) the field location, in the transition zone between the strong Mediterranean influence of the Terra Quente Transmontana and the greater continental influence of the Terra Fria Transmontana, where the vegetation is dominated by *Quercus pyrenaica* and *Quercus rotundifolia*, and where climate conditions may not always be the in the optimal growth range for cork oak; iii) the more shallow soil, as soil type has been shown to have a significant influence for cork oak growth and yield (Costa et al., 2008), combined with the very low soil boron content detected several years after the plantation (the absence of this micronutrient in the soil promotes the cessation of root elongation and the reduction of leaf expansion, affecting tree survival and imposing a limitation to growth (Camacho-Cristóbal et al., 2008)); iv) recurrent shrub encroachment episodes, namely, by *Rubus* spp., *Genista* spp. and *Cytisus* spp., that impose stronger interspecific competition for resources (e.g., light, water and soil nutrients); v) the presence of a major rodent pest, *Microtus lusitanicus*, detected especially from ages 6–7 years after planting forward.

Because the ranking of genetic effects did not change considerably between trial sites, i.e., populations with higher performance at one site remained the same at the other site, and high positive values of across-site genetic correlations were found for growth traits, it can be concluded that the significant genotype by environment interactions (G × E) found in this study occurred at a moderate level. It should be noted that our study only considered two provenance trials, and cork oak exists along a wide range of edaphoclimatic environments. Thus, further common-garden studies should be considered to improve the success of management actions in new and existing cork oak areas, particularly to assure the allocation of the most adapted seed material.

Moroccan populations showed higher growth performance at both study sites, possibly reflecting their greater investment in leaf and root traits. In fact, in a previous study conducted by Ramírez-Valiente et al. (2011), the progeny of 3 cork oak populations (PT18, ES8 and MA27) was used to study the relationship between functional traits and aboveground growth; a positive correlation was found between leaf size and growth, with the highest values of leaf size observed for plants from the Ain Rami (MA27) population. Similarly, previous results from the Monte da Fava field trial showed that among the 35 cork oak populations sampled for the genetic variation of morphological and physiological traits, Moroccan populations had higher values for leaf area (Lourenço et al., 2006). Thus, the higher photosynthetic area of Moroccan populations increases their carbon gain and, consequently, their growth potential. Lourenço et al. (2006) studied the populations at the Monte da Fava field trial 6 years after planting and verified that the Ain Rami (MA27) population had a higher water use efficiency (WUE) evaluated by isotopic signature (δ13C). Under controlled water stress conditions, Rodrigues et al. (2015) observed that this Moroccan population (MA27) also showed a more conservative water use strategy, with early stomatal closure and higher root investment when compared with other populations (ES11, FR3, IT13). These findings are likely related with the extreme rainfall regime of Ain Rami. Although the annual precipitation is 1280 mm, the drought period is very long, and summer precipitation is only 26 mm (Table 1). Coupled with its higher water use efficiency, these abilities contribute to the maintenance of a more favorable water status during drought conditions and optimized growth. The increased ability to maximize water uptake and minimize water loss may be also behind the higher survival rate of the MA28 and MA29 populations observed in our study, reflecting their capacity to endure harsher environmental conditions at the seedling stage. Indeed, a previous study found that although cork oak populations were highly vulnerable to summer drought (the main environmental constraint under Mediterranean-type climate) during the first two years after planting, seedlings originating from sites with drier summers (such as MA28 and MA29) presented higher survival rates (Ramírez-Valiente et al., 2009b).

The consistently low growth performance of the French (FR2 - Collioure and FR4 -Sartene) and Italian (IT13 - Brindisi and IT15 - Cagliari) populations was unexpected, as a previous study found that populations from the eastern part of the species' natural range, including Italian and some French populations, exhibited earlier budbursts (Sampaio et al., 2016). In fact, it would be expected that those populations would have a growth advantage, as trees can benefit earlier from favorable water and temperature conditions and experience a longer growing season overall. However, under field conditions, IT13 and FR4 were among the cork oak populations with lower water use efficiency (Lourenço et al., 2006), reflecting their greater water consumption to sustain growth. As a greater water use efficiency can be positively related to increased water stress tolerance and adaptation to drier environments (Chaves, 1991), this trait can be related to the lower (growth and survival) performance of these populations.

It should be noted that the Portuguese populations with origins close to the field trials did not show an advantage in their fitness performance. In fact, PT35, a population from the same site as the Monte da Fava field trial, displayed an intermediate survival and growth rate at this site. Additionally, PT24, a population originating from Romeu, a site close to Quinta da Nogueira, showed survival and height growth above the overall tree mean, but root collar and breast height diameter were close to the bottom ranking. These results provide evidence that local seed sources do not always have the best fitness capacity at the planting site and assisted migration may be required to assure the success of forestation. Thus, it is urgent to evaluate the way forest materials are being managed.

As cork oak occurs in a wide range of geographic and climatic conditions, it is expected that population divergence in adaptive traits may be the result of natural selection induced by environmental drivers and/or neutral evolutionary processes (including genetic drift, founder effects, and bottlenecks) (White et al., 2007). In fact, in a previous study, it was reported that the variation in functional traits observed among cork oak populations resulted from selective pressures imposed by climatic (temperature and precipitation) forces (Ramírez-Valiente et al., 2010). Our results suggest that populations from the southern and western range and from sites with warmer and drier summers performed better than the rest; therefore, it could be hypothesized that population performance of growth and survival may be related, to some extent, to local adaptation. However, as low to moderate correlations were observed between the variables of population home-sites and fitness surrogate traits, the results provide evidence that factors other than climate are likely to be relevant for cork oak adaptation.

5. Conclusion

The existence of genetic variation and plastic responses to the environment constitutes the adaptive mechanisms by which cork oak copes with changing environments. We found that a significant population variation in the survival and growth traits of cork oak and population performance seemed to be linked, to some extent, with local adaptation. Local populations had an intermediate performance while Moroccan populations, particularly MA28 and MA29, have the advantage of favoring plant establishment and improving growth, highlighting that cork oak seed origin cannot be neglected when recommending seed sources to be used in (re)forestation activities especially in a context of climate changes. Additionally, our study demonstrated that at a higher altitude, cork oak showed lower performance, likely because of environmental and soil factors. Finally, future approaches simulating tree redistribution under different scenarios should be required to consider intraspecific genetic variation. In particular, cork oak stands need more detailed and accurate predictions of response to climate change to assist management and guarantee their long-term sustainability along their existing range.

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